

CLAIMS

What is claimed is:

- 5 1. A vector system for transfection and recombinant polypeptide expression in a mammalian host cell comprising:
- (a) a first cistron encoding a transactivator protein under control of a first promoter; and
- 10 (b) a second cistron encoding an apoptosis-protective protein under the control of the first promoter or optionally under the control of a second promoter;
- wherein the first and the second cistron are contained in one or more vectors.
2. The vector system of Claim 1, further comprising a third cistron encoding at least one desired polypeptide under control of a third promoter, wherein said third promoter is responsive to the transactivator protein and wherein the first, the second, and the third
- 15 cistrons are contained in one or more vectors.
3. The vector system of Claim 2, further comprising one or more additional cistrons each encoding a desired polypeptide under control of a promoter responsive to the transactivator protein.
4. The vector system of Claim 2, wherein said polypeptide is a single chain antibody or
- 20 a heavy or light chain of an antibody or antibody fragment.
5. The vector system of Claim 1, wherein the first and second cistrons are on one vector and the first cistron lies downstream of the second cistron.
6. The vector system of Claim 1, wherein the first cistron encodes a CREB protein or a variant thereof.
- 25 7. The vector system of Claim 6, wherein the CREB protein variant is CREB variant Y134F.
8. The vector system of Claim 6, wherein the second cistron encodes an adenoviral E1b-19K protein, a Bcl-2 protein, or a Bcl-2 protein having a deletion in the regulatory loop domain.

9. The vector system of Claim 1, wherein the first cistron encodes an adenoviral E1a polypeptide or a variant thereof.

10. The vector system of Claim 9, wherein the adenoviral E1a variant comprises a mutation in CR1.

5 11. The vector system of Claim 10, wherein the adenoviral E1a variant comprises a 47H mutation.

12. The vector system of Claim 1, wherein the second cistron encodes an apoptosis-protective protein selected from the group consisting of a dominant negative mutant of p53, a protein that interacts with BAX, a protein that interacts with BAK, an inhibitor of
10 apoptosome formation, and a downstream apoptosis inhibitor.

13. The vector system of Claim 1, wherein the second cistron encodes an adenoviral E1b-19K protein, a Bcl-2 protein, or a Bcl-2 protein having a deletion in the regulatory loop domain.

14. The vector system of Claim 1, wherein the first or second promoter is an efficient
15 heterologous promoter.

15. The vector system of Claim 1, wherein the first or second promoter is a RSV-LTR promoter, a SV-40 promoter, or a cytomegalovirus promoter.

16. The vector system of Claim 2, wherein the third promoter comprises a CREB-binding element or a 19bp repeat from a hCMV-MIE enhancer.

20 17. The vector system of Claim 2, wherein the third promoter comprises a TATAA transcription initiation signal.

18. The vector system of Claim 2, wherein the third promoter is a hCMV-MIE promoter having a TATAA box region.

25 19. The vector system of Claim 2, wherein the third cistron is associated with a ubiquitous chromatin opening element, an insulator, or a barrier element.

20. The vector system of Claim 19, wherein the ubiquitous chromatin opening element comprises an extended methylation-free CpG-island.

21. The vector system of Claim 19, wherein the ubiquitous chromatin opening element comprises a hnRNP A2 promoter.

5 22. A method of expressing a desired recombinant polypeptide in a mammalian host cell comprising introducing to the mammalian host cell:

(a) a first cistron encoding a transactivator protein under control of a first promoter;

(b) a second cistron encoding an apoptosis-protective protein under control of the first promoter or optionally under control of a second promoter; and

10 (c) a third cistron encoding the desired polypeptide under control of a third promoter;

wherein said third promoter is responsive to the transactivator protein.

23. The method of Claim 22, wherein the third cistron is associated with a ubiquitous chromatin opening element, an insulator, or a barrier element.

15 24. The method of Claim 22, wherein the host cell is selected from the group consisting of a CHO cell, a mouse myeloma cell, a mouse hybridoma cell, a rat myeloma cell, and a rat hybridoma cell.

25. The method of Claim 24, wherein the host cell is a cell capable of growing in a suspension.

20 26. The method of Claim 24, wherein the host cell is a YB2/0 rat hybridoma cell.

27. The method of Claim 22, wherein the first or second promoter is an efficient heterologous promoter.

28. The method of Claim 22, wherein the transactivator and the apoptotic protective protein are homologous to the endogenous transactivator and apoptotic protective proteins
25 of the host cell.

29. The method of Claim 22, wherein the first cistron encodes a transactivator protein selected from the group consisting of an E1a protein, a CREB protein, and variants thereof.

30. The method of Claim 29, wherein the first cistron encodes CREB variant Y134F.

31. The method of Claim 22, wherein the first cistron encodes a CREB protein or a variant thereof, and the second cistron encodes a Bcl-2 protein or a Bcl-2 protein having a deletion in the regulatory loop domain.

5 32. The method of Claim 22, wherein the first cistron encodes a variant E1a protein with a mutation in CR1, and the second cistron encodes an E1b-19K protein, a Bcl-2 protein, or a Bcl-2 protein having a deletion in the regulatory loop domain.

33. The method of Claim 22, wherein the second cistron encodes an apoptosis-protective protein selected from the group consisting of a dominant negative mutant of p53,
10 a protein that interacts with BAX, a protein that interacts with BAK, an inhibitor of apoptosome formation, and a downstream apoptosis inhibitor.

34. The method of Claim 22, wherein the second cistron encodes an adenovirus E1b-19K protein, a Bcl-2 protein, or a Bcl-2 protein having a deletion in the regulatory loop domain.

15 35. The method of Claim 22, wherein said polypeptide is a single-chain antibody or a heavy or light chain of an antibody or antibody fragment.

36. The method of Claim 22, wherein said polypeptide is a part of a library of polypeptides.

37. A mammalian host cell for recombinant polypeptide expression comprising a first
20 cistron encoding a transactivator protein and a second cistron encoding an apoptosis-protective protein that prevents cell-killing due to expression of the transactivator protein.

38. The host cell of Claim 37, further comprising a third cistron encoding one or more desired polypeptide under the control of a promoter responsive to the transactivator protein.

39. The host cell of Claim 37, wherein the third cistron is associated with a ubiquitous
25 chromatin opening element, an insulator, or a barrier element.

40. The host cell of Claim 37, wherein the transactivator protein is expressed from an efficient heterologous promoter.

41. The host cell of Claim 37, wherein the first cistron encodes a CREB protein or a variant thereof, and the second cistron encodes a Bcl-2 protein or a Bcl-2 protein having a deletion in the regulatory loop domain.

42. The host cell of Claim 37, wherein the first cistron encodes a E1a variant comprising a mutation in CR1.

43. The host cell of Claim 37, wherein said cell is a CHO cell or a YB2/0 cell.

44. The host cell of Claim 37, wherein said cell is a cell capable of growing in a suspension.

45. The host cell of Claim 37, wherein said host cell is from an established cell line.

46. The host cell of Claim 37, wherein said host cell is a non-human mammalian host cell.

47. A method for producing a recombinant protein comprising culturing the host cell of Claim 37 in a suitable medium such that the one or more desired proteins are secreted into the medium.